AMENDMENTS TO THE SPECIFICATION

Please replace the paragraph beginning on page 10, line 28 with the following paragraph:

Consensus zinc finger structures may be prepared by comparing the sequences of known zinc fingers, irrespective of whether their binding domain is known. Preferably, the consensus sequence is selected from the group consisting of the consensus structure P Y K C P E C G K S F S Q K S D L V K H Q R T H T G (SEQ ID NO:8), and the consensus structure P Y K C S E C G K A F S Q K S N L T R H Q R I H T G E K P (SEQ ID NO:9).

Please replace the paragraph beginning on page 12, line 10 with the following paragraph:

A "leader" peptide may be added to the N-terminal finger. Preferably, the leader is MAEEKP (SEQ ID NO:10).

Please replace the paragraph beginning on page 28, line 1 with the following paragraph:

Another improvement to the methods of affinity screening is the control of display valency (i.e., the average number of functional zinc finger polypeptides displayed per polysome, and the capacity to vary display valency in different rounds of affinity screening. Typically, a high display valency permits many binding contacts between the polysome and nucleic acid, thus affording stable binding for polysomes which encode zinc finger polypeptide species which have relatively weak binding. Hence, a high display valency system allows screening to identify a broader diversity range of zinc finger polypeptides, since even lower affinity zinc finger polypeptides can be selected. Frequently, such low-to-medium affinity zinc finger polypeptides can be superior candidates for generating very high affinity zinc finger polypeptides, by selecting high affinity zinc finger polypeptides from a pool of mutagenised low-to-medium affinity zinc finger clones. Thus, affinity sharpening by mutagenesis and subsequent rounds of affinity selection can be used in conjunction with a broader pool of initially selected zinc finger polypeptide sequences if a high display valency method is used. Alternate rounds of high display valency screening and low display valency screening can be performed, in any order, starting from either a high or low valency system, for as many affinity screening rounds as desired, with intervening variation and sequence diversity broadening, if desired. Alternate rounds of affinity screening, wherein a first round consists of screening a zinc finger polypeptide library expressed in a high valency system, selecting zinc finger polypeptide clones which bind the target nucleic acid, optionally conducting a mutagenesis step to expand the sequence

variability of the selected zinc finger polypeptides, expressing the selected zinc finger polypeptide clones in a lower valency display system, and selecting clones which bind the ~~tarhet~~ target nucleic acid, can be performed, including various permutations and combinations of multiple screening cycles, wherein each cycle can be of a similar or different display valency. This improvement affords an overall screening program that employs systems which are compatible with switchable valency (i.e., one screening cycle can have a different display valency than the other(s), and can alternate in order).

Please replace the paragraph beginning on page 34, line 28 with the following paragraph:

This is the standard bacteriophage T4 RNA polymerase promoter, having the sequence TA ATA CGA CTA ACT ATA GGG AGA (SEQ ID NO:13).

Please replace the paragraph beginning on page 35, line 18 with the following paragraph:

Second there is a series of Ala-Ala-Val-Pro (residues 21-24 of SEQ ID NO:12) repeats. This is a standard, relatively easily translated sequence and serves to ensure that the ribosome is stalled after (and not before) the entire flexible (Ser-Gly) linker has emerged from the ribosome. This is relatively important since approximately ten amino acids are covered by the ribosome at any one time.

Please replace the paragraph beginning on page 36, line 1 with the following paragraph:

The sequence:

M	V	K	T	D	K	(SEQ ID NO:16)
ATG	GTT	AAA	ACA	GAT	AAA	(SEQ ID NO:15)

when translated, interacts with the peptidyl transferase site of the *E. coli* ribosome, causing translational pausing. In the presence of chloramphenicol, this paused state becomes a stalled state.

AMENDMENTS TO THE CLAIMS

This listing of the claims replaces all prior versions and listings:

1. (currently amended): A method for producing a zinc finger nucleic acid binding protein comprising preparing a zinc finger protein according to design rules, varying the protein at one or more positions, and selecting variants that bind to a target nucleic acid sequence by polysome display.

2. (currently amended): ~~A~~ The method according to claim 1, wherein the protein is varied at one or more positions selected from the group consisting of +1, +5, +8, -1, +2, +3, and +6.

3. (currently amended): ~~A method for producing a zinc finger nucleic acid binding protein comprising an at least partially varied sequence and selecting variants thereof which bind to a target DNA strand, comprising the steps of~~ The method of claim 1, wherein the design rules comprise:

(i) preparing a nucleic acid binding protein of the Cys2-His2 zinc finger class capable of binding to a nucleic acid triplet in a target nucleic acid sequence, wherein binding to each base of the triplet by an α-helical zinc finger nucleic acid binding motif in the protein is determined as follows:

a) if the 5' base in the triplet is G, then position +6 in the α-helix is Arg; or position +6 is Ser or Thr and position ++2 is Asp;

b) if the 5' base in the triplet is A, then position +6 in the α-helix is Gln and ++2 is not Asp;

c) if the 5' base in the triplet is T, then position +6 in the α-helix is Ser or Thr and position ++2 is Asp;

d) if the 5' base in the triple is C, then position +6 in the α-helix may be any amino acid, provided that position ++2 in the α-helix is not Asp;

e) if the central base in the triplet is G, then position +3 in the α-helix is His;

f) if the central base in the triplet is A, then position +3 in the α-helix is Asn;

g) if the central base in the triplet is T, then position +3 in the α-helix is Ala, Ser or Val; provided that if it is Ala, then one of the residues at -1 or +6 is a small residue;

h) if the central base in the triplet is C, then position +3 in the α-helix is Ser, Asp, Glu, Leu, Thr or Val;

i) if the 3' base in the triplet is G, then position -1 in the α-helix is Arg;

j) if the 3' base in the triplet is A, then position -1 in the α-helix is Gln;

k) if the 3' base in the triplet is T, then position -1 in the α-helix is Asn or Gln;

l) if the 3' base in the triplet is C, then position -1 in the α-helix is Asp;

(ii) varying the resultant polypeptide at at least one position; and

(iii) selecting the variants which bind to a target nucleic acid sequence by polysome display.

4. (currently amended): ~~A method according to any preceding claim~~ The method of claim 3, wherein the or each zinc finger has the general primary structure

(A) X^a **C** $X_{2\text{-}4}$ **C** $X_{2\text{-}3}$ F X^c X X X X L X X **H** X X X^b **H** - linker

-1 1 2 3 4 5 6 7 8 9

wherein X (including X^a, X^b and X^c) is any amino acid.

5. (currently amended): ~~A~~ The method according to claim 4 [~~5~~], wherein X^a is F/Y-X or P-F/Y-X.

6. (currently amended): ~~A~~ The method according to claim 4 ~~or claim 5~~, wherein $X_{2\text{-}4}$ is selected from any one of: S-X, E-X, K-X, T-X, P-X and R-X.

7. (currently amended): ~~A~~ The method according to claim 4 ~~any one of claims 4 to 6~~ wherein X^b is T or I.

8. (currently amended) ~~A~~ The method according to claim 4 ~~any one of claims 4 to 7~~ wherein $X_{2\text{-}3}$ is G-K-A, G-K-C, G-K-S, G-K-G, M-R-N or M-R.

9. (currently amended) ~~A~~ The method according to claim 4 ~~any one of claims 4 to 8~~ wherein the linker is T-G-E-K or T-G-E-K-P.

10. (currently amended): ~~A~~ The method according to claim 4 ~~any one of claims 4 to 9~~ wherein position +9 is R or K.

11. (currently amended): ~~A~~ The method according to claim 4 ~~any one of claims 4 to 10~~ wherein positions +1, +5 and +8 are not occupied by any one of the hydrophobic amino acids F, W or Y.

12. (currently amended): ~~A~~ The method according to claim 11 wherein positions +1, +5 and +8 are occupied by the residues K, T and Q, respectively.

13. (currently amended): ~~A~~ The method according to claim 4 ~~any preceding claim~~ wherein the polysome display technique comprises the steps of:

(a) introducing a population of mRNA species into an in vitro translation system under conditions suitable for translation to form a pool of polysomes displaying nascent zinc finger polypeptides;

(b) contacting the polysomes with a target nucleic acid under suitable binding conditions;

(c) selecting polysomes which are specifically bound to the nucleic acid; and

(d) reverse transcribing and amplifying the isolated mRNA.

REMARKS

Claims 1-13 are presently pending in the application and are allowable except for formal matters that are addressed by the foregoing amendments. In particular, the claims have been amended to correct improper multiple dependencies.

In addition, pages 11, 12, 34, 35 and 36 of the specification have been amended to insert reference to the appropriate sequence listing identifier. Furthermore, the typographical error on page 28, line 24 has also been corrected.

Submitted herewith are copies of the references not submitted with the IDS filed 14 February 2002, and a new 1449 listing these references. Applicants request these references be considered.

In view of the foregoing amendments and attached references, Applicants submit that the application is now in condition for allowance and request early notification to that effect.

Please address all correspondence to the undersigned.

Respectfully submitted,

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